

# Diameter-Related Variations of Geometrical, Mechanical, and Mass Fraction Data in the Anterior Portion of Abdominal Aortic Aneurysms

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## WHAT THIS PAPER ADDS

Irreversible dilation of abdominal aortic aneurysms (AAAs) results in larger aortic diameters. The present study investigates variations of AAA data with increased AAA diameter. Patient specific geometrical, mechanical, and biochemical data were analyzed and correlated with relevant maximum AAA diameters, which are reported for the first time in the literature. For geometrical parameters, maximum intraluminal thrombus thickness, wall thickness, and AAA expansion rate were measured and showed changes of AAA geometries for different diameters. Biomechanical investigations focus on biaxial responses and dissection properties of AAA tissues that play a key role in aneurysmal development and rupture. Biochemical tests quantify dry weight percentages of elastin and collagen within the AAA wall, which are closely related to the tissue microstructure. The present findings may advance understanding of the effects of AAA enlargement on patient specific vascular tissue properties.

**Objective:** Maximum aortic diameter is an important measure in rupture prediction of abdominal aortic aneurysms (AAAs). Analyzing the variations of geometrical, material, and biochemical properties with increased AAA diameters advances understanding of the effect of lesion enlargement on patient specific vascular properties.

**Methods:** 96 AAA samples were harvested during open surgical aneurysm repair. Geometrical factors such as the maximum intraluminal thrombus (ILT) thickness, wall thickness, and AAA expansion rate were measured. Biaxial extension and peeling tests were performed to characterize the biaxial mechanical responses and to quantify the dissection properties of aneurysmal tissue. Mass fraction analysis quantified the dry weight percentages of elastin and collagen within the AAA wall. Linear regression models were used to correlate geometrical, mechanical, and mass fraction data with maximum AAA diameter.

**Results:** Both ILT thickness and AAA expansion rate increased and were positively correlated with maximum AAA diameter, while there was a slight increase in wall thickness for AAAs with a larger maximum diameter. For the biaxial mechanical responses, mean peak stretches and maximum tangential moduli in the circumferential and longitudinal axes did not correlate with maximum AAA diameters. However, the quantified energy to propagate tissue dissections within intima-media composites showed a significant inverse correlation with maximum AAA diameter. Elastin content decreased significantly with increasing AAA diameter.

**Conclusion:** Larger AAA diameters are associated with thicker ILTs, higher AAA expansion rates, and pronounced elastin loss, and may also lead to a higher propensity for tissue dissection and aneurysm rupture.

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## INTRODUCTION

An abdominal aortic aneurysm (AAA) is a localized and irreversible dilation of the aorta.<sup>1,2</sup> The pathogenesis of

AAA, which involves a complex series of events, has not yet been fully clarified.<sup>3</sup> Continuous AAA growth may lead to wall rupture, a catastrophic event frequently associated with high mortality and serious life threatening morbidity if not addressed.<sup>4–6</sup> Currently, no technique or criterion available can provide a reliable patient specific prediction of the AAA rupture risk. Typically, patients with an AAA diameter  $\geq 5.0$  cm in women and  $\geq 5.5$  cm in men undergo surgical repair.<sup>7–9</sup> However, this “maximum diameter

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criterion" is unreliable, as indicated by previous studies.<sup>8–11</sup> In particular, it neglects the potential rupture risk for smaller AAAs.<sup>12</sup>

Rupture of an AAA occurs when the peak wall stress exceeds the local wall strength at any location. A refined understanding of the biomechanical properties of vascular tissues during AAA pathological progression may help to better explain the rupture mechanism due to tissue growth and remodeling from a biomechanical point of view. As AAA dilation is irreversible, there is a need to investigate the changes in the material properties of AAA tissues with respect to continuously increasing diameters. Such correlations re-evaluate the role of the maximum AAA diameter criterion in clinical rupture risk assessment.

The aim of the present study was to investigate how patient specific (mechanical and biochemical) properties vary with the maximum AAA diameter in patients before surgery. In particular, for individual AAA samples the maximum intraluminal thrombus, wall thicknesses and AAA expansion rate were measured and correlated with the maximum AAA diameter. The biaxial mechanical responses and the dissection properties of AAA tissue were determined quantitatively in order to investigate variations in biomechanical conditions in the aneurysm. Finally, dry weight percentages of elastin and collagen within the AAA wall were identified for different AAA diameters.

## MATERIALS AND METHODS

### Material

96 patients (mean age  $70 \pm 12$  years) who underwent open surgical aneurysm repair from November 2008 to November 2012 at the clinical department of vascular surgery, Medical University of Graz, Austria were included. Use of the material was approved by the ethics committee of the Medical University of Graz. All samples, including intraluminal thrombi (ILTs) and thrombus covered walls, were harvested from the anterior part of the aneurysm (for the particular location see the sketch in Di Martino et al.<sup>13</sup>). The samples were stored in Dulbecco's Modified Eagle's Medium within the operating room immediately after retrieval. Biomechanical analysis was performed without delay after transport to the biomechanics laboratory at Graz University of Technology. The mean duration from storage to test was  $2 \pm 1$  hours.

### Geometrical factors

**Computed tomography angiography protocol.** Computed tomography angiography (CTA) examinations were undertaken on a 128 row dual source CT scanner with the patient in the supine position during a single breath hold (SOMATOM Definition AS; Siemens Healthcare, Munich, Germany). The scanning parameters were as follows: slice thickness 2 mm, increment 1.5 mm, tube voltage 100–120 kV. For contrast enhanced CTA a bolus of 100 mL non-ionic iodinated contrast medium (Iopromidum, Ultravist 300, 300 mg iodine/mL; Bayer Schering Pharma, Berlin, Germany) was

injected intravenously (4 mL/second) via an 18 gauge catheter placed in the antecubital vein followed by 40 mL saline flush. Multiplanar reconstruction images of the abdominal aorta were routinely produced in the axial, sagittal, and coronal planes with 2 mm slice thickness. All image data sets were stored on the picture archiving and communication system.

**CTA analysis.** Dual energy data were post-processed on a workstation (Multimodality Workplace, Siemens Healthcare) running Syngo software (version VA 11; Siemens Healthcare). Diameters were measured from the outer to outer wall of the aneurysm using electronic calipers with a zoom function by a single experienced observer. The maximum diameter of the infrarenal aorta was measured as the maximal external cross sectional measurement in any plane using electronic calipers at right angles to the image slices using a set protocol on the workstation, as described in the literature.<sup>14</sup>

**AAA expansion rate.** The AAA expansion rate, defined as an increment of the maximum transverse AAA diameter per year, was obtained from consecutive CTA scans, as described above, measuring at the correlating aortic level, as identified by the neighboring anatomical structure (i.e., vertebrae, ribs). The difference between consecutive CTA examinations was divided by the interval between CT scans (minimum of 12 months) and growth described as mm/year. As the growth rate was not part of a standardized measurement and follow up protocol, no data from sequential ultrasound investigations were included.

**Wall and ILT measurements.** The maximum ILT thicknesses were measured at three different locations of the thickest part for each ILT sample and then averaged. The mean thickness of the thrombus covered wall was measured by a PC controlled video extensometer with a full image charge coupled device camera (for details of the measurement protocol see the studies by Sommer et al.<sup>15</sup> and Tong et al.<sup>16</sup>).

### Biomechanical tests

Biaxial extension tests were performed to characterize the mechanical responses of the intact thrombus covered wall at the applied (engineering) stress of  $P_{\theta\theta} = P_{LL} = 150$  kPa. Details of the specimen preparations and the experimental protocol for biaxial extension tests have been described in a previous study.<sup>17</sup>

Peeling tests quantitatively determine the energy required to propagate a dissection within a tissue.<sup>16</sup> Rectangular strips were cut with a uniform dimension of  $18.0 \times 6.0$  mm (length  $\times$  width) and further given an initial cut (incision of about 2.0–3.0 mm in length) using a surgical scalpel in order to better control the initiation of the tissue failure. Subsequently, two "tongues" were obtained and mounted onto a PC controlled, screw driven, high precision tensile testing device. The tests were executed in phosphate buffered saline solution at  $37.0 \pm 1.0$  °C and the extension

rate of 1 mm/minute was maintained throughout the test. The aneurysm wall underneath the thrombi is frequently associated with atherosclerosis, indicating that the intima and media cannot be accurately separated owing to the lack of a layer specific structure. In the current study, therefore, the peeling tests were performed on the intima–media composite and the remaining adventitia.

### Mass fraction analysis

In the present study the mass fraction analysis was based on the determination of the dry weight of elastin and collagen within the AAA intima–media and the adventitia. Tissue was frozen in liquid nitrogen and homogenized by grinding with a mortar and pestle. The wet and constant dry weight reached by drying for 3–4 days at 105 °C, of the homogenate were determined, respectively. Details regarding elastin quantitation and collagen analysis have been described in a previous study.<sup>18</sup> In brief, elastin was extracted from the dried tissue homogenate with 0.25 M oxalic acid at 100 °C for 1 hour. Elastin content was calculated from a standard curve determined by co-processing elastin standards of 12.5, 19, 25, 35, and 50 µg. Collagen was hydrolyzed by 2 M NaOH at 121 °C for 20 minutes in an autoclave. Released hydroxyproline was oxidized by chloramine-T. The content of hydroxyproline was calculated from a standard curve determined by co-processing hydroxyproline standards of 0–20 µg. Collagen content was calculated from its hydroxyproline content of 12.5%.

### Data analysis

Cauchy stress (force per current area) and stretch (ratio between final length and initial length of a material line) were computed to quantify the biaxial biomechanical responses of the tissues. The mean peak stretch (MPS) values ( $\lambda_{\theta, \max}$ ,  $\lambda_{L, \max}$ ) for the thrombus covered AAA wall in the circumferential and longitudinal axes were identified. The maximum tangential modulus (MTM), with dimension MPa, defined as the slope of the non-linear stress stretch curve at

the maximum load, was calculated and further averaged to assess stiffness at  $P_{\theta\theta} = P_{LL} = 150$  kPa of the AAA wall.

For the peeling test, the force/width values denoted by  $F_{pc}$  and  $F_{pl}$  were measured and averaged for the circumferential and longitudinal strips, respectively. Furthermore, the dissection energy per reference area, say  $W^{\text{dissect}}$ , during the (circumferential c and longitudinal l) peeling (p) was quantified by subtracting the elastic energy  $W^{\text{elastic}}$  from the external work  $W^{\text{ext}}$ , i.e.  $W_{pc}^{\text{dissect}} = (W_c^{\text{ext}} - W_{pc}^{\text{elastic}})/L_{pc}$  and  $W_{pl}^{\text{dissect}} = (W_l^{\text{ext}} - W_{pl}^{\text{elastic}})/L_{pl}$ , where  $L_{pc}$  and  $L_{pl}$  denote the reference lengths of the strips in the circumferential and longitudinal axes respectively.

All quantified data (geometrical, mechanical, mass fraction) for each variable were plotted as a function of the related maximum AAA diameter. To fit the scatter data points, a linear regression model in the optimization toolbox ('linear fit') of OriginPro 8.5 was then utilized. Correlation was evaluated with the Pearson's rank correlation coefficient  $R$ . A  $p$ -value < .05 determined statistical significance. Correlation was considered as strong for  $R \geq .40$ .

## RESULTS

During the reported period, 248 patients underwent AAA repair. Open repair was performed in 190 (76.6%) patients; endovascular repair (EVAR) was performed in only 58 (23.4%) patients owing to very conservative management protocol. A pre-operative CTA was performed in all patients. Sixty nine of 96 (71.9%) patients underwent surgery within 4 weeks of CTA and 27 within 5 months. Data for body mass index, smoking, concomitant diabetes mellitus, renal function, chronic obstructive pulmonary disease, hypertension, and AAA shape are shown in Table 1.

The resulting geometrical (maximum ILT thickness, wall thickness, AAA expansion rate), mechanical (mean peak stretch, maximum tangential modulus, dissection energy), and biochemical (mass fraction of elastin and collagen) data are plotted against the maximum AAA diameter (Figs. 1–4).

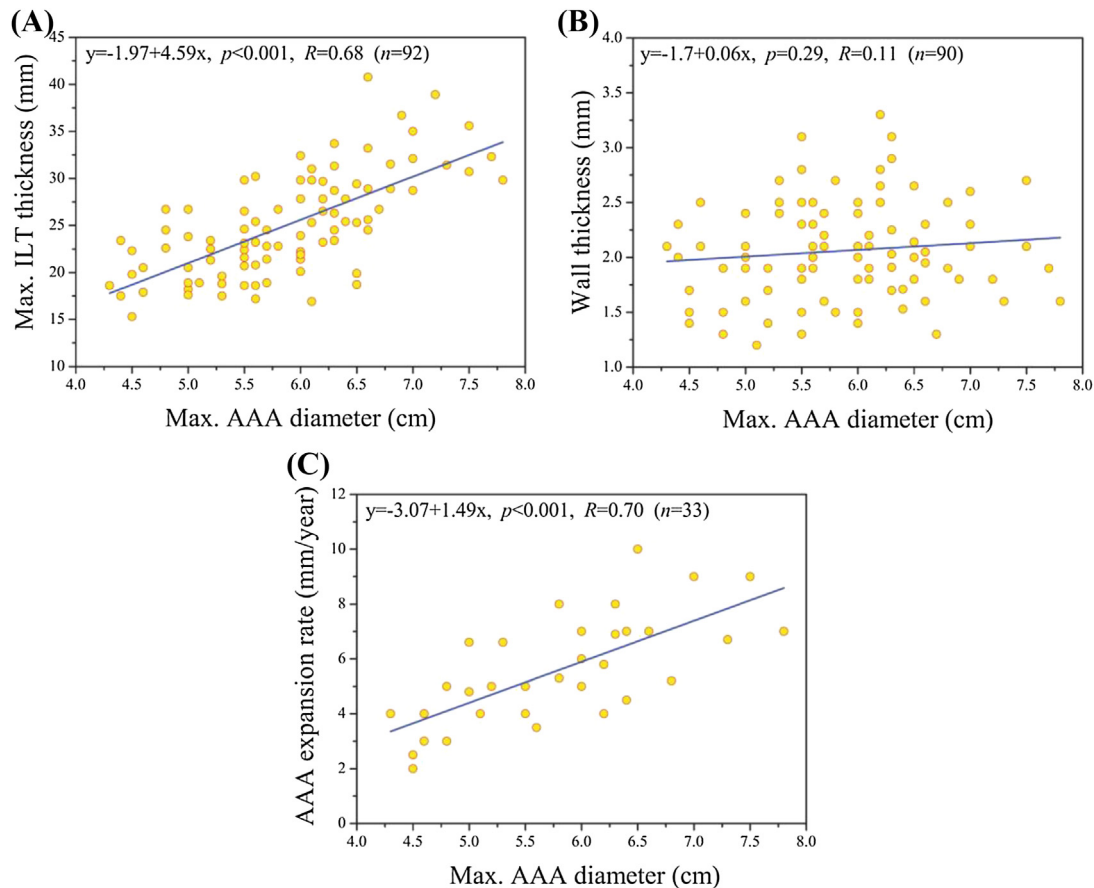
**Table 1.** Clinical data of 96 patients with abdominal aortic aneurysms (AAAs).

		Men	Women	All	Range
Clinical presentation, $n$ (%)	Asymptomatic	66	9	75 (78.1)	
	Symptomatic	13	3	16 (16.7)	
	Ruptured	5	0	5 (5.2)	
BMI (mean $\pm$ SD)		26.4 $\pm$ 3.6	26.2 $\pm$ 4.5	26.3 $\pm$ 3.7	18.1–35.2
Smoker, pack years (mean $\pm$ SD)		49.7 $\pm$ 50.8	18.8 $\pm$ 18.9	45.8 $\pm$ 49.0	4.5–170
Never smoked, $n$ (%)		10	2	12 (12.5)	
DM, $n$ (%)		13	2	15 (15.6)	
Serum creatinine, mg/dL (mean $\pm$ SD)		1.25 $\pm$ 0.62	0.92 $\pm$ 0.22	1.21 $\pm$ 0.60	0.69–4.36
COPD, $n$ (%)		12	2	14 (14.6)	
Hypertension, $n$ (%)	No hypertension	1	0	1 (1.0)	
	Controlled	55	11	66 (68.8)	
	Poorly controlled	28	1	29 (30.2)	
AAA shape, $n$ (%)	Fusiform	72	10	82 (85.4)	4.3–11.0 cm <sup>a</sup>
	Saccular	12	2	14 (14.6)	4.2–11.0 cm <sup>b</sup>

Note. BMI = body mass index; DM = diabetes mellitus; COPD = chronic obstructive pulmonary disease.

<sup>a</sup> Diameter 6.1  $\pm$  1.0 cm.

<sup>b</sup> Diameter 6.2  $\pm$  1.9 cm.



**Figure 1.** Scatter plots of the measured geometrical factors: (A) maximum intraluminal thrombus (ILT) thickness, (B) wall thickness, (C) abdominal aortic aneurysm (AAA) expansion rate with respect to the maximum AAA diameter. Predictive equations obtained by linear regression model, associated with  $p$  and  $R$ , and number,  $n$ , of specimens are shown in each plot. Both maximum ILT thickness and AAA expansion rate are significantly correlated with the maximum AAA diameter.

The related equations obtained by the linear regression model, associated with  $p$  and  $R$ , and the number of specimens,  $n$ , are provided in each figure. For each individual test, a different number of specimens of the 96 harvested AAA samples was obtained, for example only 33 intact AAA walls could be obtained for the biaxial extension tests because in several cases the AAA wall obtained from surgery was too small (and hence used for the other tests), and in some cases the mechanical test failed. It was never possible to perform biaxial extension and peeling tests from one harvested AAA wall piece. Moreover, for some AAA samples no thrombus covered wall was harvested. Some of the raw data regarding the mechanics and mass fraction have been published in previous studies.<sup>17,18</sup> However, more samples have been tested and included to provide a larger dataset for this analysis.

### Geometrical factors

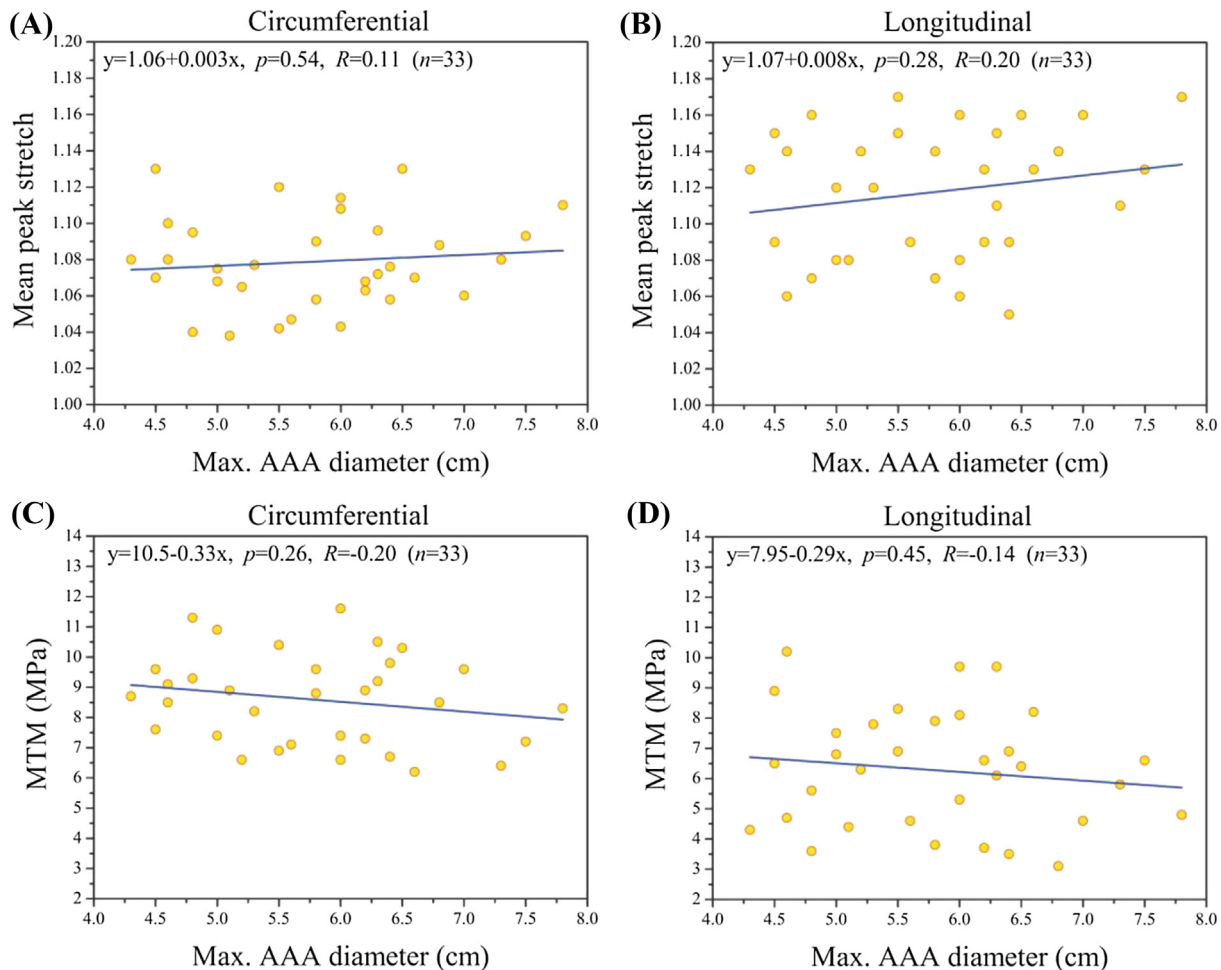
Maximum ILT thickness, wall thickness, and AAA expansion rate were measured and analyzed in 92, 90, and 33 patients, respectively. The maximum ILT thickness ranged from 15.3 to 40.8 mm (median 24.5 mm; mean  $\pm$  SD  $25.3 \pm 5.5$  mm). The wall thickness ranged from 1.2 to 3.3 mm (median 1.9 mm; mean  $\pm$  SD  $2.0 \pm 0.5$  mm). The AAA expansion rate

ranged from 2 to 10 mm/year (median 5.2 mm/year; mean  $\pm$  SD  $5.6 \pm 2.0$  mm/year). Scatter plots of the individual geometrical factors with respect to the maximum AAA diameter are shown in Fig. 1. Both maximum ILT thickness ( $p < 0.01$ ,  $R = 0.68$ ) and AAA expansion rate ( $p < 0.01$ ,  $R = 0.70$ ) were significantly correlated with the maximum AAA diameter.

### Biomechanical properties

For the biaxial extension tests, the MPS and MTM values were quantified and analyzed for 33 intact AAA walls. The MPS ranged from 1.04 to 1.13 (median 1.07; mean  $\pm$  SD  $25.3 \pm 5.5$ ) and 1.05 to 1.17 (median 1.13; mean  $\pm$  SD  $1.12 \pm 0.04$ ) in the circumferential and longitudinal axes respectively. The MTM ranged from 6.2 to 11.6 MPa (median 8.7 MPa; mean  $\pm$  SD  $8.6 \pm 1.5$  MPa) and 3.1 to 10.2 MPa (median 5.4 MPa; mean  $\pm$  SD  $6.3 \pm 2.0$  MPa) in the circumferential and longitudinal axes, respectively. Scatter plots of the mechanical quantities (MPS and MTM) with respect to the maximum AAA diameters are shown in Fig. 2. No significant correlations were found for these data with respect to the maximum AAA diameter.

For the peeling tests, the dissection energy was quantified and analyzed for 29 patients. Within the intima—media



**Figure 2.** Scatter plots of the quantified mean peak stretches (MPS) and maximum tangential moduli (MTM) in the circumferential and longitudinal axes of the intact thrombus-covered walls with respect to the maximum AAA diameter. Predictive equations obtained by linear regression model, associated with  $p$  and  $R$ , and number,  $n$ , of specimens are shown in each plot; no significant correlations between the biaxial mechanical quantities and the maximum AAA diameter.

(I + M) composite, the dissection energy ranged from 3.2 to 7.1 (median 5.0; mean  $\pm$  SD  $5.3 \pm 1.2$ ) and 4.2 to 9.7 (median 6.4; mean  $\pm$  SD  $6.6 \pm 1.3$ ) in the circumferential and longitudinal axes, respectively. Within the adventitia, the dissection energy ranged from 6.8 to 11.8 (median 9.7; mean  $\pm$  SD  $9.3 \pm 1.5$ ) and 5.3 to 10.5 (median 8.4; mean  $\pm$  SD  $8.5 \pm 1.1$ ) in the circumferential and longitudinal axes, respectively. Scatter plots of the quantified dissection energy with respect to the maximum AAA diameters are shown in Fig. 3. The dissection energy within the intima—media composite was significantly inversely correlated with the maximum AAA diameter (circumferential:  $p = 0.01$ ,  $R = 0.46$ ; longitudinal:  $p = 0.02$ ,  $R = 0.44$ ).

### Mass fraction

Mass fractions of elastin and collagen were quantified in 23 AAA samples. Dry weight percentages of elastin within the intima—media composite and the adventitia ranged from 3.1% to 11.7% (median 6.4%; mean  $\pm$  SD  $6.8 \pm 2.5$ ) and 7.2% to 13.1% (median 9%; mean  $\pm$  SD  $9.4 \pm 1.7$ ), respectively. Dry weight percentages of collagen within the

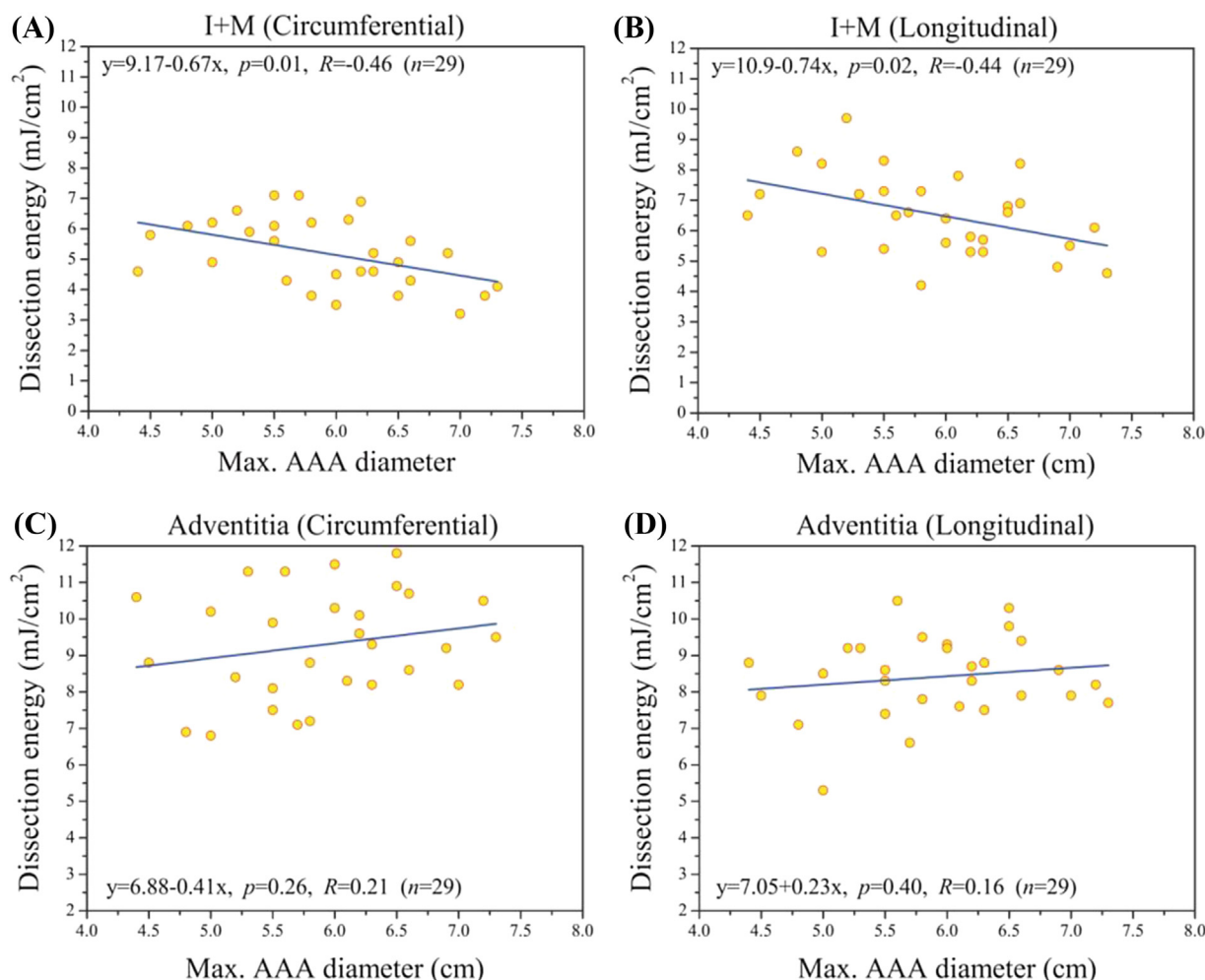
intima—media composite and the adventitia ranged from 16.6% to 28.9% (median 21%; mean  $\pm$  SD  $22.3 \pm 3.5$ ) and 12.5% to 22.5% (median 17.2%; mean  $\pm$  SD  $17.5 \pm 2.6$ ), respectively. Scatter plots of the quantified dissection energy with respect to the maximum AAA diameters are shown in Fig. 4. The mass content of elastin within the wall was significantly decreased and significantly correlated with the AAA maximum diameter (intima-media:  $p < 0.01$ ,  $R = -0.66$ ; adventitia:  $p = 0.02$ ,  $R = -0.51$ ).

### DISCUSSION

Aneurysm development is a process associated with permanent aortic dilation. Despite the available criteria to predict the rupture risk, the maximum AAA diameter is an important measure for examining AAA progression. In the present study variations in geometrical, mechanical, and biochemical data of aneurysmal tissues were measured with respect to the related maximum AAA diameter.

The maximum ILT thickness was significantly correlated with maximum AAA diameter (see Fig. 1A), indicating a pronounced ILT thickening with aneurysm enlargement. It

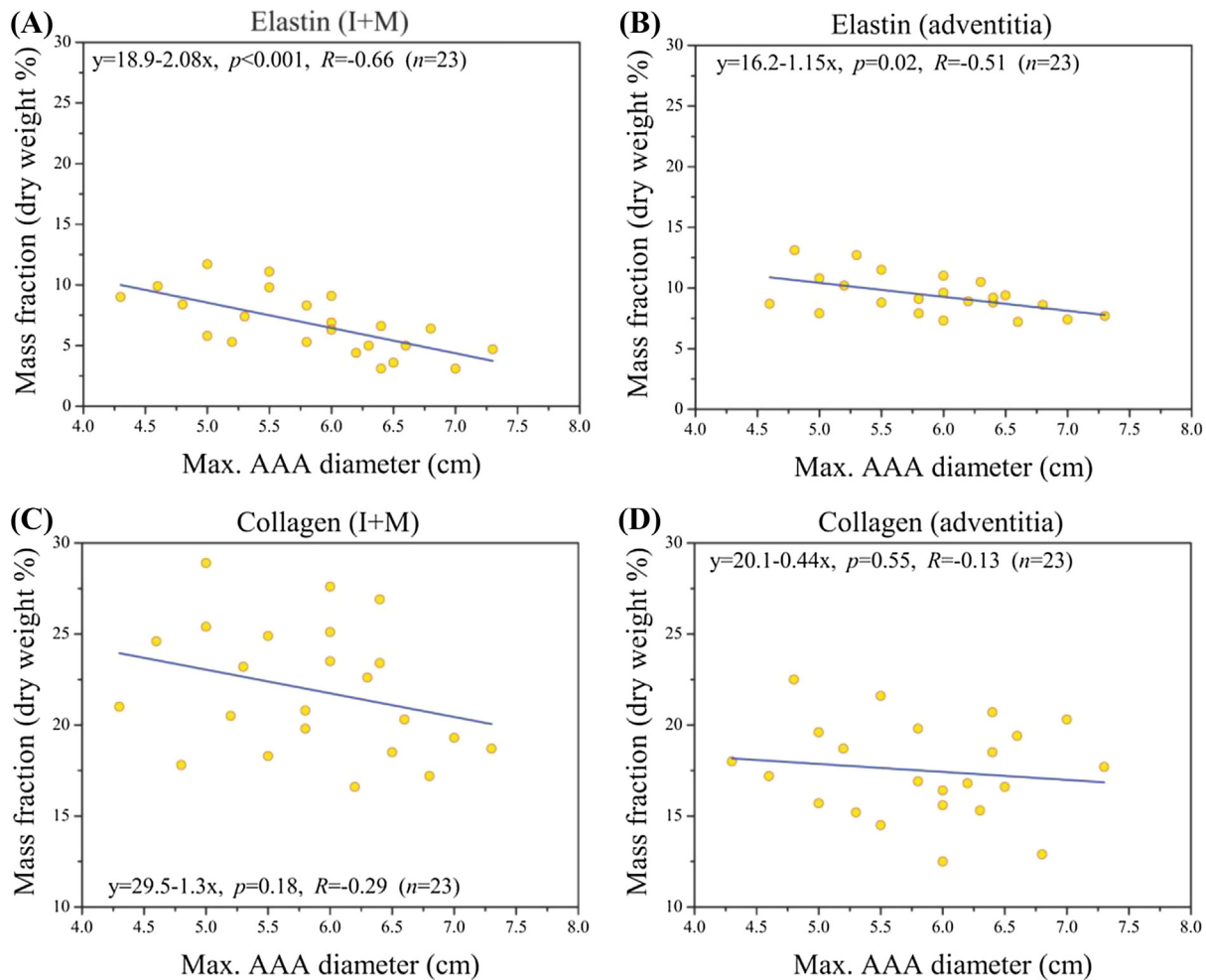




**Figure 3.** Scatter plots of the quantified energy to dissect the intima—media (I + M) and the adventitia in the circumferential and longitudinal axes with respect to the maximum AAA diameter. Predictive equations obtained by linear regression model, associated with  $p$  and  $R$ , and number,  $n$ , of specimens are shown for each plot. Dissection energy within the intima—media composite is significantly inversely correlated with the AAA maximum diameter.

has been suggested that ILT is associated with wall weakening. The underlying pathomechanism has not yet been clearly identified. Reduced oxygen transport from the flowing blood to the AAA wall leading to hypoxia induced cell dysfunction has been discussed,<sup>19</sup> as well as granulocyte activation acting on metalloproteinase activation.<sup>20</sup> Therefore, a larger transverse AAA diameter may lead to a more severe wall weakening and to a lower wall strength. However, as a mechanical cushion, the thicker ILT may also reduce/alter the wall stress distribution in that region.<sup>21,22</sup> Therefore, a larger AAA diameter associated with a thicker ILT cannot simply predict the rupture risk for the specific patient AAA. The wall thickness, as shown in Fig. 1B, increased slightly, possibly owing to atherosclerotic intimal thickening, inflammatory processes and/or calcification,<sup>23,24</sup> but it was not significantly correlated with the maximum AAA diameter. It should be emphasized that the wall thickness is not a critical rupture related factor, and an *in vivo* rupture does not necessarily occur at the thinnest wall region.<sup>25</sup> In addition, regional distribution of the wall thickness requires some future effort to investigate how

the thrombus free wall thickness varies with AAA enlargement.<sup>26</sup> The AAA expansion rate has long been linked to factors such as ILT volume and wall strength in the literature.<sup>22,27–30</sup> In the current study, a positive correlation between the AAA expansion rate and the maximum AAA diameter was found (see Fig. 1C). Note that each data point shown in Fig. 1C represents the averaged expansion rate of one patient during the time interval between the first inspection and surgery (which could be weeks, months, or years) with respect to its maximum diameter before surgery. These data points do not represent the expansion rates at a specific diameter value for one patient. A higher expansion rate represents a more significant enlargement of the AAA over a time period, which is, to a great extent caused by elastin degradation and collagen turnover. More importantly, a rapid collagen remodeling due to cessation of new collagen biosynthesis could result in aneurysm rupture.<sup>31,32</sup> Hence, the higher expansion rate for patients with the larger diameter AAAs should be considered as a key factor in AAA rupture risk assessment.



**Figure 4.** Scatter plots of the quantified mass fractions for elastin and collagen within the intima-media (I + M) and the adventitia with respect to the maximum AAA diameter. Predictive equations obtained by linear regression model, associated with  $p$  and  $R$ , and number,  $n$ , of specimens are shown for each plot. Elastin content within the intima-media composite and the adventitia are significantly correlated with the AAA maximum diameter.

As can be seen from Fig. 2, there was a slight increase of MPS and a relatively lower MTM for AAAs with larger maximum diameters. However, both mechanical results in the circumferential and longitudinal axes, were not significantly correlated with the maximum AAA diameter. This suggests that the extensibility and the tissue stiffness of the aneurysm wall in response to the same stresses did not change significantly for AAAs with larger maximum diameters. Therefore, an increased AAA diameter is not necessarily accompanied by wall degeneration in terms of biaxial mechanical responses. Note also that the applied stress level did not cause tissue rupture when biaxially stretched. To measure the ultimate tensile strength the uniaxial tensile test is the method of choice.

For the peeling tests, the quantified dissection energy within the intima-media (I + M) composite was significantly correlated with the maximum AAA diameter (see Fig. 3A, B). This suggests that a higher propensity for tissue dissection may exist for AAAs with larger maximum diameters. One of the consequences caused by tissue dissection within the intima-media is to alter the biomechanical conditions within the AAA, which may lead to wall rupture due to an imbalance

between peak wall stress and wall strength. As shown in Fig. 3C and D, the dissection properties of the adventitia did not vary significantly as the AAA enlarged.

Mass fraction analyses suggest that the dry weight of elastin within both the intima-media (I + M) and the adventitia is significantly decreased for larger diameter AAAs (see Fig. 4A, B). The elastin loss contributes to an increased aortic wall distensibility,<sup>28</sup> and is regarded as the main trigger for aneurysm formation. A substantial reduction in elastin within the AAA wall, when compared with non-aneurysmal aorta, has been reported in several previous studies.<sup>31,33,34</sup> Here biochemical investigations focus on the variations in the elastin content (in layer specific aortic structure) with the progressively enlarged AAA diameter after aneurysm formation. A continuous decrease in elastin in this process emphasizes its key role in tissue growth and remodeling during wall degeneration. Owing to lack of mass fraction data for thrombus free walls, a quantitative comparison is not available to show whether the pronounced elastin loss in AAA growth is affected by ILT or not.

Despite a slight decrease, the mass content of the collagen within the AAA wall was not significantly

correlated with the maximum AAA diameter (see Fig. 4C, D). Compared with healthy aortas, collagen cross links in the aneurysmal aortic wall are increased owing to continuous accumulation of aged collagen.<sup>31</sup> Meanwhile, biosynthesis could be reduced, limiting the new collagen production, which would lead to a decrease in mass content. Moreover, the collagen turnover promotes lesion enlargement and is closely related to the mechanical properties of the AAA wall. In this regard, an important microstructural parameter is the collagen fiber distribution with respect to changes in AAA diameter, not investigated in the present study.

Finally, some limitations of this study are summarized. First, the number of AAA samples considered for the *in vitro* experiments was relatively low. Second, the findings in the aortic tissue do not necessarily reflect the nature of a specific aneurysm wall. The quantified geometry and material properties relate only to each AAA at the time of surgery. Third, all AAA diameter measurements were performed on axial CT planes, and therefore can overestimate the actual diameter. Additionally, as AAA growth occurs in a non-linear fashion,<sup>3,35,36</sup> growth determination by retrospective evaluation of CT examinations is a limitation in the study protocol. Finally, all samples were harvested from the anterior portion of the aneurysm sac. However, most AAA ruptures occur on the lateral or posterior aspect.<sup>37,38</sup> Excision of the posterior aneurysm sac increases the risk of intra- and post-operative bleeding, and was not considered for this study. Therefore, this analysis may not be representative of the true biological process involved in lateral or posterior AAA expansion and rupture, because of the heterogeneous nature of the aortic wall and the specific changes at the point of rupture.<sup>39</sup>

## CONCLUSION

These data indicate that the maximum ILT thickness and the averaged AAA expansion rate are significantly increased for larger AAA diameters. The quantified energy to dissect within aneurysmal intima-media composites is significantly inversely correlated with the maximum AAA diameter. Moreover, a decrease in elastin content is identified and significantly correlated with the increased AAA diameter. Therefore, it is concluded that a larger maximum AAA diameter is associated with a thicker ILT, a higher AAA expansion rate, a pronounced elastin loss, and a higher propensity for tissue dissection.

## CONFLICT OF INTEREST

None.

## FUNDING

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